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## **Acute and short-term effects of the nonpeptide endothelin-1 receptor antagonist bosentan in humans**

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**Abstract:** In recent years, evidence from various animal experiments has accumulated that emphasizes the role of endothelin-1 in the pathophysiology of several cardiovascular diseases, including congestive heart failure. The recent advent of potent antagonists of this system now allows the assessment of the involvement of endothelin-1 in the maintenance of vascular tone in animals and humans. We report hemodynamic data from two trials in patients with chronic severe congestive heart failure (i.e., reduced left ventricular ejection fraction of <30%, elevated resting pulmonary capillary wedged pressure >15 mmHg, and/or reduced cardiac index of 2.5 L/min/m<sup>2</sup> or less) who were treated with the mixed endothelin-type A and type B-receptor antagonist bosentan. In the first study, the acute effect of bosentan (300 mg, intravenous) on hemodynamics and neurohormones was investigated. Bosentan was well tolerated and significantly improved impaired hemodynamics due to systemic and venous vasodilation. In the second trial, bosentan was given orally (0.5 g bid) for 14 days, in addition to conventional triple treatment for congestive heart failure, including digitalis, angiotensin-converting enzyme inhibitors, and diuretics. Cardiac hemodynamics were monitored during the first 24 hours of treatment, and measurements were repeated during the last day of bosentan therapy. Bosentan was well tolerated in these patients as well, and hemodynamic measures were compatible with an additional effect of bosentan after 2 weeks. However, there was a slight increase in heart rate as well. Our result underline the importance of endogenously generated endothelin-1 in congestive heart failure and suggest a potential benefit of endothelin antagonism in such patients. However, long-term studies are needed to establish whether chronic endothelin antagonism has beneficial clinical effects and is capable of improving survival and/or symptoms in severe heart failure patients who remain symptomatic despite standard triple therapy

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## Acute and Short-Term Effects of the Nonpeptide Endothelin-1 Receptor Antagonist Bosentan in Humans

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**Summary.** In recent years, evidence from various animal experiments has accumulated that emphasizes the role of endothelin-1 in the pathophysiology of several cardiovascular diseases, including congestive heart failure. The recent advent of potent antagonists of this system now allows the assessment of the involvement of endothelin-1 in the maintenance of vascular tone in animals and humans. We report hemodynamic data from two trials in patients with chronic severe congestive heart failure (i.e., reduced left ventricular ejection fraction of <30%, elevated resting pulmonary capillary wedged pressure >15 mmHg, and/or reduced cardiac index of 2.5 L/min/m<sup>2</sup> or less) who were treated with the mixed endothelin-type A and type B-receptor antagonist bosentan. In the first study, the acute effect of bosentan (300 mg, intravenous) on hemodynamics and neurohormones was investigated. Bosentan was well tolerated and significantly improved impaired hemodynamics due to systemic and venous vasodilation. In the second trial, bosentan was given orally (0.5 g bid) for 14 days, in addition to conventional triple treatment for congestive heart failure, including digitalis, angiotensin-converting enzyme inhibitors, and diuretics. Cardiac hemodynamics were monitored during the first 24 hours of treatment, and measurements were repeated during the last day of bosentan therapy. Bosentan was well tolerated in these patients as well, and hemodynamic measures were compatible with an additional effect of bosentan after 2 weeks. However, there was a slight increase in heart rate as well. Our result underline the importance of endogenously generated endothelin-1 in congestive heart failure and suggest a potential benefit of endothelin antagonism in such patients. However, long-term studies are needed to establish whether chronic endothelin antagonism has beneficial clinical effects and is capable of improving survival and/or symptoms in severe heart failure patients who remain symptomatic despite standard triple therapy.

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**Key Words.** endothelin-1, ET<sub>A/B</sub>-receptor antagonism, human heart failure, hemodynamics

Intense laboratory and clinical research over the last 15 years has unequivocally documented that the endothelium is one of the major sources of substances involved in the regulation of vascular muscle tone and, thus, vascular resistance by releasing both vasodilator

and vasoconstrictor mediators [1–3]. While the function of endothelium-derived relaxing factor, presumed to be nitric oxide [4], in the regulation of arteriolar tone in normal subjects [5,6] and in many diseases, among them chronic heart failure [7], hypertension [8], hypercholesterolemia [9], and smokers [6], is well established the role of the vasoconstrictor peptide endothelin-1 [2] is much less clear. Evidence is accumulating that endothelin-1 is abuminally released and is directed towards the vessel walls, where it increases vascular muscle tone, acting as an autocrine or paracrine stimulator. The pharmacological effects of infused endothelin-1 are well recognized [10]. Besides being a functionally important vasoconstrictor, it is involved in the regulation of electrolyte balance and modulates secretion of other hormonal systems that regulate mitogenesis and growth-related metabolism [11–13].

Increased plasma concentrations of this potent vasoconstrictor have been described in many circumstances, such as renal failure [14], severe hypertension [15], pulmonary hypertension [16], coronary endothelial dysfunction [17], coronary vasospasm [18], and chronic heart failure [19,20] and may be related to a poor prognosis after myocardial infarction [21]. Similarly, in severe heart failure, high levels of plasma big endothelin-1, the precursor of endothelin-1, are associated with low survival and seem to predict 1-year mortality better than hemodynamic variables or neurohormonal prognostic markers of congestive heart failure [22]. However, these findings obviously do not prove an involvement of endothelin-1 in the pathophysiology of these conditions. Rather, they might reflect reduced renal and/or pulmonary clearance or endothelial damage with increased peptide release. Likewise, the finding that endothelin-1

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infusions resulting in pathophysiological blood concentrations caused profound vasoconstriction in animals [23] and, similarly, following big endothelin-1 infusion in humans [24], is suggestive for a role of endothelin-1 in many diseases associated with high circulating endothelin-1 levels but does not provide clearcut proof.

The recent development of specific endothelin-receptor antagonists [25] now provides the means to clarify the role of endothelin-1 in the physiological and pathophysiological regulation of vascular tone in humans. Importantly, it was shown that administration of an endothelin-type A receptor antagonist, BQ 123, caused profound forearm vasodilation when infused directly into the brachial arteries of normal volunteers [26], but the endothelin-receptor antagonist bosentan did not have hemodynamic effect in normal, anesthetized dogs [27]. Because patients with heart failure often have elevated levels of plasma endothelin-1, we decided to use one of these endothelin-receptor antagonists, bosentan, in such patients, assessing changes of systemic hemodynamics as a means to further elucidate the importance of this system in the maintenance of increased vascular tone that characterizes this condition. In addition, possible links between this endothelial regulator of vascular tone and the renin-angiotensin-aldosterone system will be discussed.

### ***Endothelin-1, its Receptors, and Bosentan***

Endothelin-1 is a 21-amino acid peptide with potent and sustained vasoconstrictor properties. It is converted from the precursor plasma big endothelin-1 by an inhibitable metalloprotease. To date, two endothelin receptor subtypes have been identified, the endothelin-type A (ET<sub>A</sub>) and type B (ET<sub>B</sub>) receptors. While the ET<sub>A</sub> receptor is predominantly involved in endothelin-1-mediated vasoconstriction, the ET<sub>B</sub> receptors might play a dual role, that is, induce vasoconstriction in some tissues [28] and vasodilatation in certain other tissues [29]. Bosentan is a competitive, selective, and mixed endothelin receptor antagonist that specifically blocks both ET<sub>A</sub> and ET<sub>B</sub> receptors sites [25] and is orally active. Simultaneous systemic blockade of both receptor types was judged to be of advantage when examining the effect of acute and short-term inhibition of the hemodynamic and humoral consequences caused by the increased endothelin-1 levels.

### ***Acute Hemodynamic Effects of Bosentan in Severe Chronic Heart Failure***

#### ***Patients***

Two female and 22 male patients (35–70 years, mean  $51.9 \pm 10$  years) with congestive heart failure of more

than 3 months duration and dyspnea according to NYHA class III were studied [30]. All patients had a left ventricular ejection fraction of  $<30\%$  (mean  $23.2 \pm 4.4\%$ ) and pulmonary artery wedge pressure was  $>15$  mmHg and/or cardiac index at rest reduced ( $<2.5$  l/min/m<sup>2</sup>).

All patients were clinically stable. Angiotensin-converting enzyme inhibitors were discontinued for four plasma half-lives prior to study for safety reasons because this was the first administration of this compound in such patients. Other medications were withheld on the morning of the study, with the exception of antiarrhythmic drugs ( $n = 4$ ).

#### ***Methods***

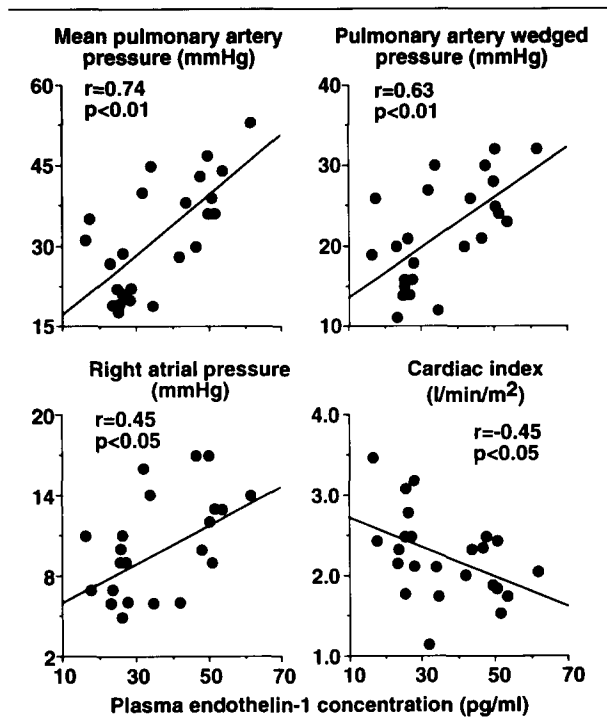
Systemic hemodynamics were measured invasively using standard techniques, using flow-guided thermodilution catheters for the measurement of right sided cardiac and pulmonary artery pressures and cardiac output. Blood pressure was measured directly from the radial or brachial artery and heart rate from the electrocardiogram.

Patients were randomized to receive double-blind placebo or bosentan. Treatment was started with intravenous infusion of 100 mg of bosentan or matching placebo (0.9% saline) over 15 minutes. Sixty minutes after starting the first infusion, an additional 200 mg of bosentan or placebo was infused in an identical manner. The cumulative dosage of 300 mg of bosentan infusion was assumed to reach plasma concentrations that were comparable with those obtained in animals studies with experimental congestive heart failure in which endothelin antagonism was shown to be effective. As an additional safety measure, the infusions were performed in two dose steps, separated by a washout period sufficient to allow for hemodynamic "adjustment."

Hemodynamic measurements were obtained 5, 20, 40, and 60 minutes after the onset of each bosentan infusion. Blood for hormone determination (endothelin-1, big endothelin-1, noradrenaline, plasma renin activity) was drawn immediately before bosentan infusion and 60 minutes after the first and second infusions, respectively. Differences in hemodynamic and hormonal measurements in the two study groups were compared using analysis of variance for repeated measures. Finally, plasma concentrations of bosentan were determined. Mean maximum plasma concentrations of bosentan reached  $10 \pm 3$  µg/mL 20 minutes after the first and  $32 \pm 8$  µg/mL 20 minutes after the second infusion, respectively.

#### ***Results***

Baseline hemodynamic values were not significantly different in both study groups. Expectedly, though, plasma endothelin-1 levels were elevated in these patients [ $36.4 \pm 13.7$  (SD) pg/ml; normal  $<15$  pg/ml] and, as described earlier [31], were directly correlated with mean pulmonary artery, pulmonary artery



**Fig. 1.** Relationships between baseline plasma endothelin-1 concentrations and the extent of pulmonary hypertension, cardiac filling pressures, and cardiac index in patients with chronic heart failure. The correlations indicate that higher plasma endothelin-1 concentrations are associated with a greater degree of pulmonary hypertension, more severely elevated right- and left-sided cardiac filling pressures, and lower cardiac index.

wedge pressure, and right atrial pressures, and indirectly with cardiac index (Fig. 1). This finding indicates that the severity of hemodynamic impairment is related to the activation of this system, as measured by endothelin-1 levels in blood, but it has to be kept in mind that the main action of this system occurs presumably in an autocrine/paracrine fashion in the vessel wall [32].

The hemodynamic effects of randomized administration of either placebo or bosentan are shown in Figures 2 and 3, which depict absolute changes from baseline measurements. Placebo infusion did not induce significant hemodynamic changes throughout the study period, attesting to the hemodynamic stability of the patients and the feasibility of the experimental procedure. In contrast, bosentan significantly reduced mean arterial and pulmonary artery pressure, and right atrial and pulmonary artery wedge pressures, and increased cardiac and stroke volume index, while heart rate did not change. Consequently, calculated systemic vascular resistance decreased significantly, as did pulmonary vascular resistance. As illustrated in Figures 2 and 3, an initial marked effect of bosentan was seen only 20 minutes after the first (100-mg) infu-

sion, and this effect persisted for up to 60 minutes. Infusion of the second dose in addition to the first dose appeared to have an additional effect, at least on measured pressures, but it is difficult if not impossible to say whether this effect is due to the additional dose and, possibly, more complete endothelin-receptor blockade or whether it represents the continued and increasing effect of the first bosentan dose.

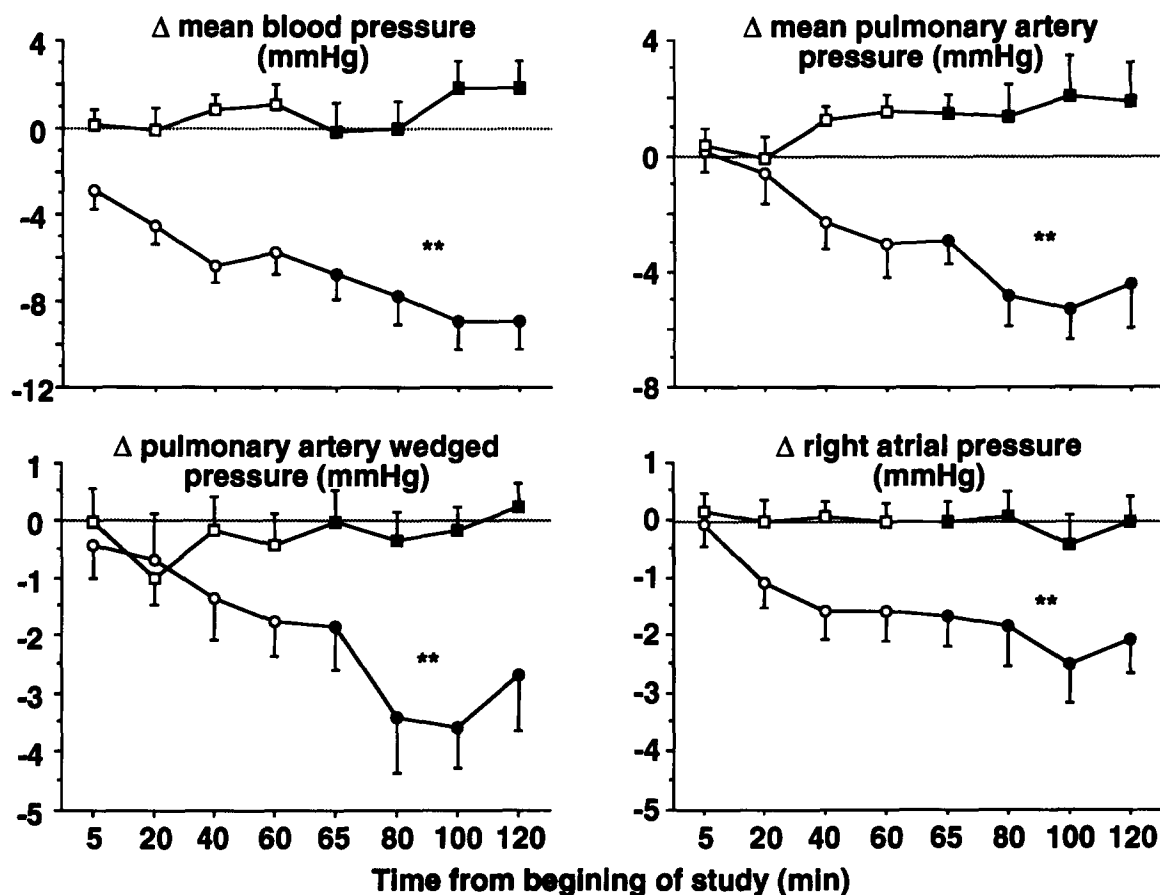
Plasma endothelin-1 levels were comparably elevated in both study groups at baseline and increased significantly in bosentan-treated patients (from  $38.9 \pm 11.4$  to  $89.6 \pm 27.1$  pg/ml,  $p < 0.01$ ) and remained unchanged in placebo-treated patients. Plasma big endothelin-1, norepinephrine, angiotensin II, and renin activity remained unchanged in both groups. None of the measured hormones was significantly correlated with hemodynamic changes.

### Limitations and potential implications

Taken together, the study for the first time provides evidence that acute systemic treatment with an endothelin receptor antagonist results in significant hemodynamic changes in patients with chronic heart failure and elevated circulating endothelin-1 levels, demonstrating that this endothelial vasoconstrictor system plays a role in the circulatory pathophysiology in these patients. Therefore, interference with this system by either specific receptor antagonists or with endothelin-1 converting enzyme inhibitors may not only prove useful for the further delineation of this system's role in circulatory physiology and pathophysiology, but may lead to a new therapeutic approach. The hemodynamic profile of systemic, venous, and pulmonary arterial vasodilation and unchanged heart rate, as seen in this study, appears to be compatible, at least on theoretical grounds, with a beneficial role of chronic endothelin-1 antagonism in patients with heart failure. However, a number of points need comment.

The present study cannot answer whether blockade of circulating endothelin-1 effects or paracrine/autocrine effects induced by abnormally released endothelin-1 accounts for the observed hemodynamic changes. Both explanations appear possible because endothelin-1 infusions resulting in concentrations of twice the normal range had pronounced hemodynamic and antinatriuretic effects in animals [23]. Thus, the endothelin-1 system might prove to play a dual role in the control of vascular function by both local tissue and circulatory components, the relative importance of which will have to be determined.

Plasma endothelin-1 levels, but not those of its precursor big endothelin-1, increased more than twofold after bosentan, a finding similar to that seen after acute administration in dogs [27] or chronic oral therapy in mineralocorticoid-induced hypertension in rats [33]. We cannot readily explain this phenomenon, but displacement of endothelin-1 from its receptor sites or a decreased clearance of endothelin-1 due to  $ET_B$



**Fig. 2.** Changes of arterial, pulmonary artery, pulmonary artery wedge, and right atrial pressures in patients with chronic heart failure given intravenous placebo or the endothelin-receptor antagonist bosentan. Placebo (squares) did not produce significant hemodynamic effects. In contrast, bosentan (circles) significantly lowered all measured pressures (\*\*  $p < 0.01$  by repeated-measures ANOVA). The greater portion of the response was seen with the lower bosentan dose [100 mg iv (open circles)], but the higher dose [200 mg iv (closed circles)] seemed to have an additional effect.

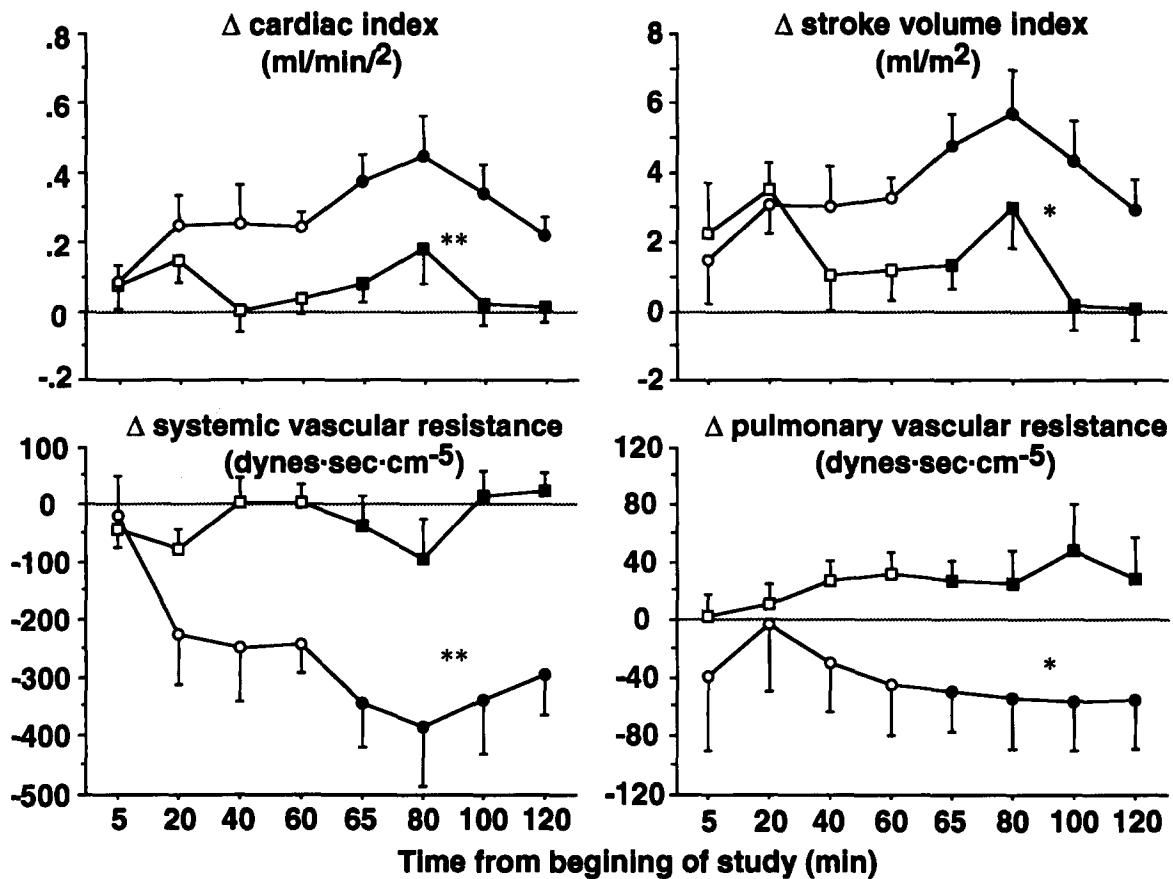
receptor blockade appears possible [34]. Increased synthesis subsequent to interruption of a negative feedback mechanism also cannot be excluded, but big endothelin-1 did not increase, rendering this explanation unlikely. Increased conversion of big endothelin-1 into endothelin-1 might also explain the finding, but a reduction of big endothelin-1 would then be expected. Clearly, more data are needed to assess the significance and underlying mechanisms of this finding.

Exogenous endothelin-1 has heterogeneous vascular effects, with a greater renal than coronary action in animals [23] and a doubling of renal vascular resistance as compared with an increase of mean blood pressure of only approximately 25% as found in humans when big endothelin-1 was infused [24]. The measurement of systemic hemodynamic parameters, as in this study, can only give an integrated view of the possibly different responses of various vascular beds, and it remains to be seen whether endothelin

antagonism in humans also has more marked effects in some vascular beds as compared with others. Thus, it will be of importance to see whether endothelin-1 antagonism would also influence endothelin-1-induced antinatriuresis [23], a phenomenon of great potential importance, particularly in chronic heart failure.

The results from this acute hemodynamic study certainly do not indicate either beneficial hemodynamic effects during chronic therapy or improvement of symptoms, such as increased exercise tolerance or reduced dyspnea. So far, we even do not know which patients might respond acutely to this kind of therapy. Animal data suggest that bosentan has little if any effect in rats with mild degrees of heart failure and that endothelin-1 antagonism is increasingly effective when heart failure becomes more severe and plasma endothelin-1 levels are elevated [35].

Plasma endothelin concentrations also increase in patients with more severe degrees of heart failure



**Fig. 3.** Changes of cardiac and stroke volume index, and of systemic and pulmonary vascular resistance, in patients with chronic heart failure given intravenous placebo or the endothelin-receptor antagonist bosentan. Placebo (squares) did not produce significant hemodynamic effects. In contrast, bosentan (circles) significantly increased cardiac and stroke volume index and lowered systemic and pulmonary vascular resistance (\*\*  $p < 0.01$  by repeated-measures ANOVA). The greater portion of the response was seen with the lower bosentan dose [100 mg iv (open circles)], while the effect of the higher dose [(200 mg iv (closed circles))] appeared to be less consistent on these parameters as compared with its effects on pressure measurements.

[20], and our results in patients with moderate to severe heart failure confirm previous findings of relationships between plasma endothelin-1 levels and the extent of pulmonary hypertension, left ventricular filling pressure, and indices of systolic dysfunction, for example, cardiac output or left ventricular ejection fraction [19,20]. Thus, patients with more severe heart failure and elevated plasma endothelin-1 levels, as in this study might, on theoretical grounds, respond better to endothelin receptor antagonist therapy than patients with less hemodynamic impairment. We did not find a relationship between either endothelin-1 or big endothelin-1 plasma concentrations and hemodynamic changes after bosentan. However, the number of patients was too small to ascertain the predictive value of circulating endothelin-1 and big endothelin-1 levels with respect to hemodynamic effects after blockade of its vascular effects.

### Hemodynamic Effects of Short-Term Oral Endothelin-Receptor Antagonist Therapy

#### Rationale and study patients

Studies using a rat model of chronic heart failure have shown that the effect of bosentan was additive to that of an angiotensin-converting enzyme inhibitor [35]. For safety reasons, angiotensin-converting enzyme inhibitors were discontinued in the acute study of bosentan, and, therefore, no conclusions could be drawn regarding the safety and possible additional hemodynamic effects of combined angiotensin-converting enzyme inhibitor and endothelin-receptor antagonist therapy in patients with heart failure. Because additional effects might be of benefit in a number of patients who remain symptomatic on angiotensin-

converting enzyme inhibitors, we performed a pilot trial to assess both the safety and the hemodynamic effects of oral bosentan in six men with chronic heart failure and dyspnea, NYHA class III, despite treatment with an angiotensin-converting enzyme inhibitor, digoxin, and diuretics. All patients had a left ventricular ejection fraction of  $<30\%$  and pulmonary artery wedge pressure was  $>15$  mmHg in the morning before taking their medications.

### **Study design and methods**

Patients were studied hemodynamically twice 14 days apart using the same techniques as described earlier. On the first study day, 500 mg of bosentan was added in the morning and in the evening to the regular therapy, and hemodynamic measurements were obtained over 24 hours following the morning dose. Patients were then discharged on their regular therapy, to which bosentan 500 mg bid orally was added in an open fashion, and patients were restudied in an identical manner 14 days later. Statistical analysis was performed comparing hemodynamics at days 1 and 14 using analysis of variance for repeated measures.

### **Results**

Bosentan added to the patients' established therapy with diuretics, digoxin, and an angiotensin-converting enzyme inhibitor was well tolerated, and there were no untoward biochemical effects in serum electrolytes, creatinine, or liver enzymes. The main hemodynamic findings are depicted in Figure 4. When interpreting the results from this study, it is important to recall that due to the study design and the nature of the study being primarily a safety study, no conclusions can be drawn about the additional hemodynamic effects of bosentan on the first study day. It is evident, however, that therapy reduced systemic vascular resistance and blood pressure, and the cardiac index increased. Changes in mean blood pressure were identical during day 14 [mean difference throughout all measurements, 95% CI,  $-2.6$  ( $0.35$  to  $-5.5$ ) mmHg], but systemic vascular resistance was lower at day 14 [mean decrease  $-232$  ( $-122$  to  $-342$ ) dynes  $\text{sec}/\text{cm}^5$ ]. Cardiac index was increased [mean increase  $0.35$  ( $0.60$  to  $-0.10$ )  $1/\text{min}/\text{m}^2$ ], but there was also an increase in heart rate [mean increase  $6.5$  ( $2.7$  to  $10.2$ ) beats/min]. Stroke volume was numerically higher throughout day 14 as compared with day 1 [mean increase  $2.6$  ( $6.3$  to  $-2.1$ )  $\text{ml}/\text{m}^2$ ], but this effect was not significant, as were changes observed for right atrial, pulmonary artery, or pulmonary capillary wedge pressures.

### **Conclusions**

The conclusions that can be drawn from this study are obviously limited. However, it is important to note that additional therapy with bosentan was well tolerated, and there was no symptomatic hypotension. The difference in hemodynamic parameters after 14 days

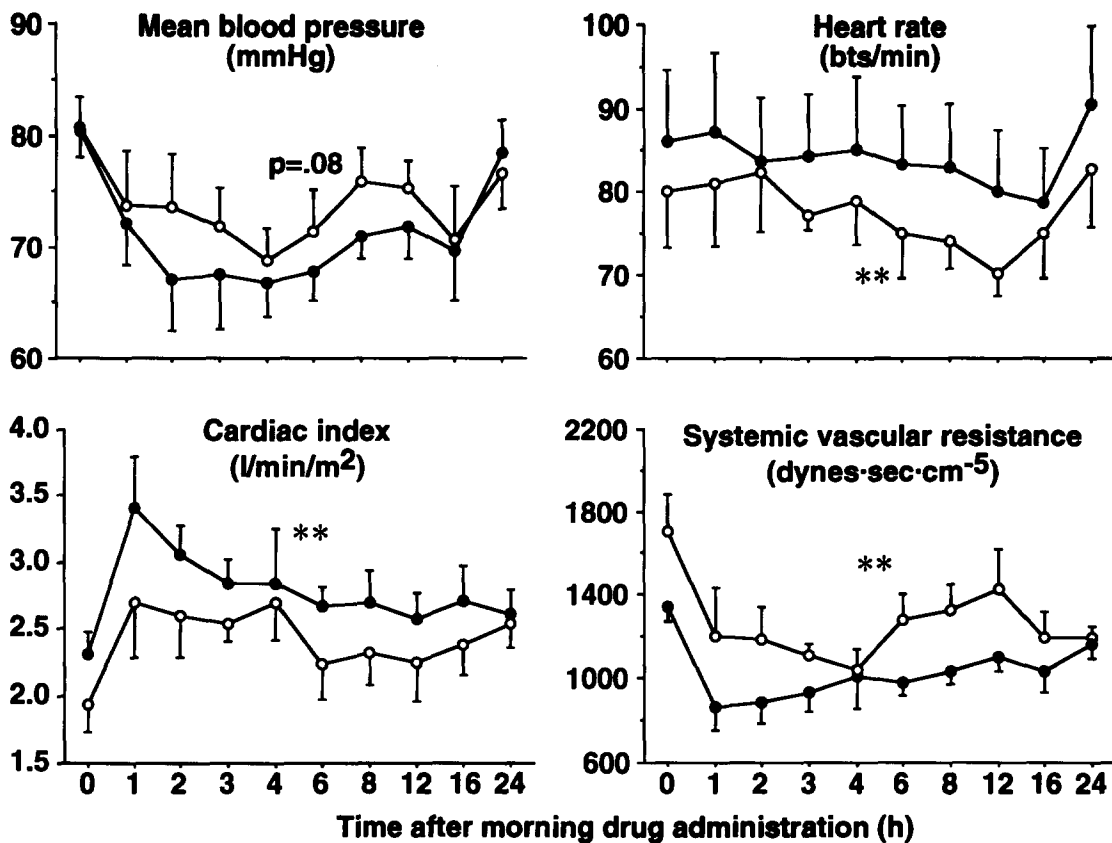
of oral therapy would be compatible with a greater chronic than acute effect, but results from placebo-controlled trials will have to be awaited to clarify this issue. The study also cannot answer the question of what would constitute an appropriate dose. There is no way, thus far, to test the completeness of receptor blockade of this paracrine/autocrine system, and dose finding will have to rely on surrogate endpoints, for example, hemodynamic changes or improvement of exercise tolerance time.

It is also worthy to note that heart rate was higher during day 14. This finding, which could indicate sympathetic activation due to arterial vasodilation and baroreceptor stimulation, differs from the lack of heart-rate effects of acute bosentan administration [30]. Interestingly, the pattern of heart rate throughout both study days appeared to be similar, but it is obvious that the important issue regarding possible sympathetic activation during sustained endothelin-receptor antagonist therapy clearly needs further study.

### **Effects Beyond Hemodynamic Benefit?**

The preliminary data obtained in patients with heart failure thus far seem to be encouraging enough to further pursue studies of endothelin-receptor antagonists, particularly in patients not adequately treated with angiotensin-converting enzyme inhibitors. However, it will also be of importance to see whether effects other than those related to the regulation of vascular tone can be demonstrated by pharmacological interference with this system.

Endothelin-1 has been shown to exert powerful mitogenic effects, either directly or indirectly, in a variety of cells, including cardiac myocytes [12,36], vascular smooth muscle cells [11], mesangial cells [37], and cardiac fibroblasts [38]. Interestingly, endothelin-1 seems to be involved in the mediation of the mitogenic effects of angiotensin II, thus pointing to a potentially important interaction between these two systems in chronic heart failure. Both endothelin-1 and angiotensin II significantly stimulated [ $^3\text{H}$ ]-thymidine incorporation into cardiac fibroblasts, whose effects were dose-dependently inhibited by an  $\text{ET}_A$  receptor antagonist (BQ123) [38], and angiotensin II-mediated vascular smooth muscle cell growth can be partly explained by angiotensin II-induced endothelin-1 production [39]. Also, in cultured rat cardiomyocytes, angiotensin II upregulated the cardiac endothelin-1 gene in the same manner as did the immediate-early protooncogenes, and the late induction of the  $\text{ET}_B$  receptor, mainly via the cardiac angiotensin II type I receptor, is compatible with an involvement of endogenous endothelin-1 in angiotensin II-induced cardiac hypertrophy [40]. Angiotensin II also upregulates  $\text{ET}_A$  receptors in human vascular smooth muscle cells,



**Fig. 4.** Changes in mean blood pressure, heart rate, cardiac index, and systemic vascular resistance on the first day of combined therapy with diuretics, digoxin, angiotensin-converting enzyme inhibitors, and the endothelin-receptor antagonist bosentan (open circles) and on the 14th day of combined therapy (closed circles). Bosentan 500 mg bid was given together with the other drugs in the morning and 12 hours later. \*\*  $p < 0.01$  by profile analysis of repeated measurements.

providing further evidence for a link between the renin-angiotensin and the endothelin systems [41]. Differences in the release of endothelin-1 following endothelial angiotensin II type I receptor stimulation may also be responsible for the heterogeneity in vascular smooth muscle responsiveness to angiotensin II in different vascular beds [42].

Endothelin-1 also seems to interact with the renin-angiotensin system and sympathetic nervous system. Thus, endothelin-1 stimulated the conversion of angiotensin I to angiotensin II in cultured pulmonary artery endothelial cells, thereby possibly indirectly regulating vascular tone [43], and subpressor doses of endothelin-1 potentiated the vasoconstrictor response to norepinephrine in vitro [44]. Finally, angiotensin-converting enzyme inhibitors reduced the stimulated release of endothelin-1 from human endothelial cells in culture, presumably via potentiation of autacoid formation from the cells, thereby pointing to the possibility that angiotensin-converting enzyme inhibitors may exert part of their hemodynamic and antiproliferative effects via reduction of endothelin-1 production/release and/or production of bradykinin and endothelium-derived nitric oxide [45].

Although the information regarding interactions between the renin-angiotensin and the endothelin systems so far is almost entirely based upon in vitro experiments and deductions from such experiments to the situation in humans in vivo have to be made with caution, it is interesting to speculate that combined blockade of both systems may be associated with many potentially beneficial cellular effects that clearly go beyond the hemodynamic effects that we are in the process of unraveling at present.

### Future of Endothelin Receptor Blockade

A direct comparison of the effects of  $ET_A$ -selective and mixed  $ET_A$  and  $ET_B$  receptor antagonists will be needed to determine the importance of  $ET_B$  receptor blockade in humans. This might be of particular relevance in congestive heart failure, where vasoconstriction through the vascular smooth muscle  $ET_B$ -receptor mechanism seems to be enhanced [46]. Finally, the role of downregulation or upregulation of the  $ET_A$  and/or  $ET_B$  receptors seen in association with



elevated endothelin-1 plasma levels in various experimental and animal models of congestive heart failure needs confirmation in human heart failure.

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